

Project Progress

The broad goal of this project is to investigate the ecological, physiological and genetic aspects of desiccation and freezing tolerance in cyanobacterial crusts and cryptoendolithic communities. The aim of the initial phase of this research project was to describe and identify the significant members of cryptoendolithic photosynthetic communities found in travertine terraces (calcium carbonate) originally deposited by hot springs now within Yellowstone National Park. Samples were collected from travertine formations of varying age, from as recent as 10 years ago to sites dating back over 300,000 years. In all cases, a cryptoendolithic community was present in the form of a 1–2-mm-thick greenish band just below the surface of the rock, which was composed primarily of cyanobacteria. Our hypothesis was that the species diversity of these photosynthetic communities would be low due to the extreme nature of this environment, which undergoes summer desiccation and winter freezing. Both molecular and culture based methods were used to investigate cryptoendolithic communities as well as microbiotic soil crusts in Yellowstone National Park. In the second year of this project we finished the molecular analysis of these communities, continued culture isolation efforts, and began work on physiological aspects of desiccation and freezing tolerance. From phylogenetic analysis we have learned that desiccation and freezing tolerance are distributed throughout the cyanobacterial radiation. Cultivated cyanobacterial strains from travertine communities were used in experiments on desiccation and freezing tolerance. Of the strains tested, 54% were able to recover from desiccation at 17% relative humidity, and 80% recovered from freezing at -15°C . The majority of strains were also able to tolerate salinities up to 24‰, probably a result of the overlap of adaptations for desiccation and osmotic stress. In addition to cold temperatures, 60% of the strains were able to recover from exposure to temperatures up to 45°C in the desiccated state, although they could not tolerate high temperatures in liquid culture.

Highlights

- Continued culture isolation efforts have resulted in the isolation of over 100 cyanobacterial strains from cryptoendolithic travertine environments. Comparison between sequences of culture isolates and

those of an environmental clone library suggest that we have isolated many of the ecologically relevant strains.

- Many new strains of cyanobacteria were isolated by enrichment culture from samples collected 2–3 years previously and kept under conditions of low humidity, demonstrating a high survival rate from desiccation.
- Phylogenetic trees were constructed to examine the relationships among the cultures and clone sequences within this study as well as among other cultured cyanobacteria. The overwhelming result from phylogenetic analysis is that desiccation/freezing tolerances are traits that are distributed in many cyanobacterial lineages.
- In almost all cases the nearest relative of clone library sequences from travertine communities was an uncultivated cyanobacterial sequence from wide ranging environments that included desert, Antarctic and other cryptoendolithic communities.
- Of the travertine cyanobacterial strains tested, 54% were able to recover from desiccation at 17% relative humidity, and 80% recovered from freezing at -15°C .
- In addition to cold temperatures, 60% of the strains were able to recover from exposure to temperatures up to 45°C in the desiccated state, although they could not tolerate high temperatures in liquid culture.

Roadmap Objectives

- **Objective No. 4.1:** Earth's early biosphere
- **Objective No. 5.3:** Biochemical adaptation to extreme environments

Field Expeditions

Field Trip Name: Yellowstone Field Trip

<i>Start Date:</i> 06/30/03	<i>End Date:</i> 7/18/03
<i>Continent:</i> North America	<i>Country:</i> USA
<i>State/Province:</i> Wyoming	<i>Nearest City/Town:</i> West Yellowstone, MT
<i>Latitude:</i> 44 N	<i>Longitude:</i> 111 W
<i>Name of site(cave, mine, e.g.):</i> Yellowstone National Park	<i>Keywords:</i> desiccation, cyanobacteria, microbial ecology, cryptoendolithic communities

Description of Work: Collection of travertine samples containing cryptoendolithic communities for culture isolation and molecular work.

Members Involved: